PERGAMON

Food and Chemical Toxicology 38 (2000) 385–393

Food and Chemical Toxicology

www.elsevier.com/locate/foodchemtox

Review

Allergenicity of Refined Vegetable Oils

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(Accepted 20 September 1999)

Summary—Several commercially important refined vegetable oils are derived from plants which are recognized as potent food allergens (e.g. peanut, soy). Full refining of oils results in the almost complete removal from oils of protein, which is responsible for allergic reactions. However, it is uncertain whether the minute amounts remaining could provoke allergic reactions in highly susceptible individuals. This has led to a vigorous debate about the safety of refined oils and specifically whether to label each oil individually because of the potential risk of allergenicity. Peanut oil has been the most thoroughly studied. It has been shown, in well-designed studies, that refined peanut oil can be safely consumed by the vast majority of peanut-allergic individuals, whereas unrefined oil can provoke reactions in some of the same individuals. However, some other studies report cases of allergic individuals reacting to oils, which are presumed to be refined. While it is likely that the discrepancy between these observations is due to differences in the processing of the oils, and possibly the protein content, this has not been formally demonstrated. Few data exist on the potential allergenicity of other edible vegetable oils; what data there are suggest that the major oils (soy, maize, sunflower, palm) do not provoke allergic reactions in susceptible individuals. Determining the content and immunoreactivity of the residual protein of refined oils is crucial to assessing the allergenic risk they present. Current methodology is inadequate and has not been validated for use with oils and aqueous extracts from oils. Little is known about the importance of different processing steps on allergenicity, although this information is crucial to risk assessment, particularly when considering process modifications. Available data suggest that the protein content of crude oils is of the order of 100–300 μg and that refining results in levels up to about 100-fold lower. The review concludes that peanut oil, and by extrapolation other edible vegetable oils, presents no risk of provoking allergic reactions in the overwhelming majority of susceptible people. However, there is a need to standardize and validate methodology for measuring the protein content and immunoreactivity of such so that they can be used to maintain process specifications. Thresholds of reactivity to allergens in man also need to be established in order to assess fully the risk from very small amounts.

Keywords: residual protein; threshold of reactivity; analytical methodology.

Abbreviations: BCA = bicinchoninic acid; DBPCFC = double blind, placebo-controlled food challenge.

Introduction

Refined vegetable oils are used in a wide variety of foodstuffs, and the actual type of oil used at any one time in a particular time may change because of factors such as availability. Currently, vegetable oils are labelled generically when used in this manner ('Vegetable oil'). However, several commercially important oils are derived from plants which are recognized as potent food allergens (e.g. peanut—*Arachis hypogea*, soy—*Glycine max*). As a result, there is a vigorous debate about whether to label each oil individually on the grounds of allergenic risk.

Food allergy involves almost exclusively the protein component of foods. These proteins are capable of producing severe reactions in some individuals allergic to them after ingestion, in some instances, of minute quantities of the offending allergen. Edible oils are produced from a botanically diverse range of plant species. Many of the seeds used to produce vegetable oils contain proteins which are highly allergenic. Production involves pressing the oilseed, followed by a series of processes to refine the oil to the desired degree. Although full refining of oils results in the almost complete removal of protein, thresholds of reactiv-

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0278-6915/00/$ - see front matter
PII S0278-6915(99)00158-1

ity have not been established, even for allergens that have been well studied. In these circumstances, it is plausible that sometimes enough protein remains in an oil to provoke a reaction in a highly sensitive individual. As a result, doubts have been cast on the safety of oils derived from these sources for allergic individuals, particularly in the light of well-documented cases of reactions after ingestion of some oils.

The purpose of this paper is to review the evidence on the allergenicity of edible oils and to draw conclusions regarding their safety for allergic individuals. Epidemiological evidence on adverse reactions to oils, and both published evidence on the protein content of oils as well as more recent, unpublished, preliminary data on the role of different refining steps in removing protein from the oil are considered below.

Food allergy

Mechanisms and signs

The term food allergy refers to the condition in which an immunological reaction to a foodstuff produces clinical symptoms when the affected individual eats that food. This implies that some component of the food, almost always a protein, is recognized by cells of the immune system and an immune response occurs. As with all immune responses, the initial contact with the food allergen (which may take place before birth) does not usually produce any overt reaction, but primes the immune system. This results in sensitization of the affected individual. A proportion of sensitized individuals subsequently develops allergy to the food.

Allergy to foods displays all the fundamental characteristics of other immune responses, for example vaccinations. It is antigen-specific, exhibits memory and repeated exposure usually increases the strength and speed of the response. Allergic reactions to foods can also be triggered by relatively small amounts of allergen. Although a range of immunological mechanisms have been implicated in food allergy, those which cause most concern and which appear to be most frequent are mediated by IgE, the antibody responsible for anaphylaxis in man.

The clinical manifestations of IgE-mediated allergy to foods are extremely diverse. They may involve the skin (urticaria), the respiratory system (asthma), and the oral mucosa (swelling, redness) as well as the gastrointestinal tract (nausea, vomiting, pain and discomfort). Reactions can occur minutes to hours after exposure to the allergen. In the most severe reactions, anaphylactic shock is manifested by peripheral vasodilatation that leads to unconsciousness and can rapidly result in death if untreated. A second severe, and potentially fatal, manifestation is Quincke's oedema, the typical signs are angio-oedema, laryngeal/pharyngeal oedema.

Thresholds of reactivity

Knowledge of the amount of allergen that can provoke a reaction is crucial to assessing the risk from allergens and thereby inform decisions on manufacturing practices and labelling. However, little is known about thresholds of reactivity to food allergens, even common ones, and the distribution of such thresholds among the population of allergic individuals. Knowledge of thresholds for other IgE-mediated allergic reactions (e.g. the diagnostic skin prick test) suggests that they will range over several orders of magnitude of concentration. Thus, Oppenheimer *et al.* (1992) reported that the sensitivity of a group of 11 peanut-allergic patients, as expressed in terms of skin prick test reactivity, varied over a 10,000-fold range of concentrations. However, they also showed that the lowest amounts needed to trigger a clinical reaction during a double blind, placebo-controlled food challenge (DBPCFC) spanned a narrower range (30 mg to 8 g, i.e. 267-fold). Evidence in the clinical literature on other allergens indicates that such a wide range is not atypical; a milk-allergic infant was reported to react to as little as 1 μg of bovine serum albumin (Goldman *et al.*, 1963), but generally much larger amounts are required to trigger a reaction. Work by the National Food Agency of Sweden, which investigates all serious food allergic reactions and attempts to quantify the amounts that produce reactions, has provided useful information (Foucard *et al.*, 1997). Thus, they showed that as little as 10 mg of ovalbumin was responsible for an adverse, although not fatal reaction, while 60 mg of casein was fatal in one individual. They had no data for peanuts, considered one of the most potent food allergens, since they are not commonly consumed in Sweden. A recent DBPCFC study with individuals previously found to be highly sensitive to peanut protein, showed that as little as 100 μg could produce a reaction, albeit a subjective and mild one (Hourihane *et al.*, 1997b).

Clinical reports and epidemiology of non-refined and refined vegetable oil allergenicity

Allergic reactions attributable to oils are few and most relate to peanut oil. Epidemiological studies of food allergy are still few and none have investigated the prevalence of reactivity to oils *per se*. Several clinical trials have studied reactions to oil, although most lacked statistical power. Thus, most evidence regarding oil allergenicity comes from clinical reports of individual cases and so provides an unsatisfactory basis for estimating the risk to the population at large.

Peanut oil

Peanuts (*Arachis hypogea*) are now recognized as being among the most potent allergenic foods, based both on the prevalence of peanut allergy and the frequency of reported severe adverse reactions (Loza and Brostoff, 1995; Hourihane *et al*., 1997a; Tariq *et al*., 1996). It is therefore unsurprising that peanut oil has been the most studied. Moneret-Vautrin *et al*. (1991) provided the first well-documented report which indicated that peanut oil could provoke immediate allergic reactions. They extended this initial observation by a report of four cases where peanut oil was shown to exacerbate an eczematous rash (Moneret-Vautrin *et al*., 1994). In that study, they also suggested that peanut allergens were 'more reactive' when presented in an oily matrix. In a subsequent investigation, the same group reported positive DBPCFC with 5 ml of oil in four peanut-allergic patients (Olszewski *et al*., 1998). However, they did not clearly specify whether the oil used was refined or not, nor did they give a detailed description of the challenge procedures (cited in Moneret-Vautrin,1992). In a separate study, Moneret-Vautrin *et al*. (1998) observed positive DBPCFC responses in five patients to 5 ml doses of peanut oil, presumably refined. This number represented a significant minority of the peanut-allergic individuals tested. The authors contrasted their results with those of Hourihane *et al*. (1997a), and speculated that the refining process may well underlie the observed differences between the two studies.

Where a possible role for peanut oil in the aetiology of peanut allergy has been identified, the evidence is conflicting. De Montis *et al*. (1995) observed no correlation between the frequency of peanut sensitization among infants and the presence of peanut oil in the formula milks which they consumed. However, in the same paper, they reported that the odds ratio for peanut sensitization were significantly higher in infants who had received vitamin D in peanut oil than in controls who had not. The odds ratio was further increased in those infants who had received vitamin D immediately after birth compared to those where administration had been delayed (8.25 *vs* 4.82). These data could indicate, as suggested by the authors, that residual peanut protein in the oil sensitized infants. However, an alternative interpretation could be that bolus doses, particularly in oil, which may delay absorption, were more effective at boosting existing sensitivity than doses of protein received *ad lib*. over a long period of time (as in formula milk). Unfortunately no analytical data were provided on the protein content of the oils used for the vitamin D preparation. Guéant *et al*. (1995) demonstrated that extracts and micellar preparations of peanut oil could release histamine from blood leucocytes of sensitized individuals. In the same experiment, they observed that histamine was also released from several leucocyte samples on exposure to preparations made from sunflower, soy and rapeseed oil. These findings were taken to indicate the presence of cross-reactive proteins in these oils, but no details were given about foods to which the cell donors were allergic, apart from peanut.

In contrast to the above reports, two clinical trials attest to the absence of reactivity to peanut oil among peanut-allergic individuals (Hourihane *et al*., 1997a; Taylor *et al*., 1981). In the first trial, 10 subjects with a history of peanut allergy or anaphylaxis were given up to 16 ml of refined peanut oil in capsules, using a DBPCFC procedure. No subject reacted to the oil. However, because of the small number of subjects, the trial lacked adequate statistical power. Thus, it can be shown that if the incidence of reactivity among peanut-allergic individuals to the level of peanut protein in refined peanut oil is 5%, then the probability of picking a panel which did not contain at least one individual capable of reacting was about 40%. In addition, the use of capsules would also prevent the detection of reactions from contact with the oral mucosa, a frequent observation among peanut-allergic individuals. Growing concerns about those inadequacies, particularly following the demonstration of clinical reactivity to the oil by Moneret-Vautrin *et al*., led to the second trial. 60 peanut-allergic individuals were challenged with either crude or refined peanut oil in a randomized double-blind crossover study (Hourihane *et al*., 1997b). Up to 16 ml of oil was administered in foods, suitably flavoured to mask their taste and smell. No subject reacted to the refined oil, while the crude oil provoked reactions in six subjects. This study thus demonstrated with 95% certainty that fewer than 5% of peanut-sensitive individuals would react to the amount of protein present in up to 16 ml of the refined oil. Consequently, refined peanut oil does not present a risk to the vast majority of peanut-allergic individuals. This conclusion was also reached by the a subcommittee on peanut allergy of the UK Committee on the Toxicity of Chemicals in Food, Consumer Products and the Environment following a detailed review of the literature (UK DoH, 1998).

Soybean oil

Soybean oil, despite its widespread use and the awareness of allergy to soy (*Glycine max*) products by medical practitioners, only features in a few cases of anaphylactic shock. These are of doubtful relevance to food allergy since they followed intravenous infusion of an emulsion containing soybean oil (Andersen and Nissen, 1993; Hiyama *et al*., 1989). In contrast, a study by Bush *et al*. (1985) found that three different types of soybean oil were unable to elicit reactions in people with well-documented reactions to soy by ingestion. This study has been criticized from the statistical point of view

because only seven subjects were tested. The methodology used (capsules) has also been criticized because it avoids allergen contact with the oral mucosa, a route of exposure which is now known to be of considerable importance in eliciting reactions (Ortolani *et al.*, 1993). However, it does provide at least strong circumstantial evidence against any significant allergenicity of these oils. Significantly, one of the soybean oils tested included a cold-pressed one which would be expected to have contained a relatively high amount of soy protein. Further circumstantial evidence comes from the work of the Swedish National Food Administration. Since 1994, the Administration has been recording and investigating all cases of fatal and severe reactions to foods (Foucard *et al.*, 1997). While soy featured in about 25% of the reported cases (cf 33% for peanuts), none implicated soybean oil, or a product containing soybean oil as the only source of soy.

Sunflower seed oil

Sunflower seed (*Helianthus annuus*) is infrequently reported as an allergen, with only episodic clinical reports in the literature (Axelsson *et al.*, 1994; Iwaya *et al.*, 1994; Möller and Paul, 1996; Noyes *et al.*, 1979). Sunflower oil was implicated in two reports (Kanny *et al.*, 1994; Olszewski *et al.*, 1998). In one, the response observed after a single blind oral challenge was unusual as symptoms began after 2.25 hours and developed fully after 8 hours (Kanny *et al.*, 1994). In contrast to this report, Halsey *et al.* (1986) observed that sunflower seed oil (16 ml) failed to provoke any reaction in two patients who had previously experienced anaphylactic reactions to sunflower seed.

Other oils

Allergic reactions to sesame seed (*Sesamum indicum*) are well documented, although there are no good epidemiological data (Blamoutier and Denimal, 1992; James *et al.*, 1991; Kagi and Wüthrich, 1993; Kanny *et al.*, 1996; Malish *et al.*, 1981; Steurich, 1989). However, sesame oil differs from the oils previously discussed, inasmuch as it is used principally for its flavour. It is therefore not refined and would be expected to contain significantly more protein. However, only one instance of an allergic reaction related to ingestion of the oil has been reported (Chiu and Haydik, 1991). These authors detailed a case of anaphylaxis following ingestion of sesame oil.

Couturier *et al.* (1994) reported a single instance of coconut (*Cocos nucifera*) oil allergy, apparently resulting from a 'maternalized' infant formula. Teuber *et al.* (1997b) described two cases of anaphylaxis precipitated by ingestion of coconut. They attributed sensitization in these cases to cross-reactivity with walnut.

No instances of allergic reactions to the other major edible oils, including corn (maize) (*Zea mays*) oil, palm (*Elaeis guineesis*) oil and palm kernel oil, have been reported. Several reasons may account for this. The first is that the proteins from those seeds may be intrinsically less allergenic than those of other oils. There is, however, no experimental evidence either for or against this view. The proteins from those sources may be less widely consumed by humans in the parts of the world where food allergy is likely to be diagnosed. This is probably the case for palm fruit and coconut, although maize is used extensively. Finally, the economics of handling oils from those sources may mean that they are produced by relatively few large refiners, ensuring a higher and more consistent quality of oil.

Protein content of vegetable oils

Ascertaining the protein content of oils is of vital importance for several reasons:

- The protein component is responsible for any potential allergic properties and quantification is therefore crucial to risk assessment.
- Knowledge of the effect of different refining steps on protein content can help to pinpoint the most important ones and is therefore vital to the risk assessment of new processes.
- Information about the protein content of oils used in clinical studies can help to establish thresholds of reactivity and therefore contribute to the safety of these products.

Analytical methodology

Total protein analyses. No validated methods currently exist for measuring the total protein content of edible oils. Current 'Total protein' methodology used has been as follows:

- Extraction of the oil with different buffer/aqueous systems under various conditions. The protein content can then be determined using one of several assays, for example Bradford (1976), where the dye Coomassie Blue binds specific amino acids; Lowry *et al.* (1951), which relies on aromatic amino acids or bicinchoninic acid (BCA) (Smith *et al.*, 1985), where the peptide bonds are detected by the Biuret reaction followed by dye binding. Generally, the detection is spectrophotometric for all these methods. All suffer problems with extraction efficiency and/or false positives.
- Total nitrogen analysis directly in the oil using an analyser with a boat injection module. All bound nitrogen compounds are converted to nitric oxide by high temperature oxidation. The nitric oxide formed (NO) is mixed with ozone to produce excited nitrogen oxide (NO_2*). The light excited

by the NO_2* molecules is a measure for the amount of bound N in the sample. This method may overestimate the protein content, since nitrogen from all sources will be measured (e.g. phospholipids as well proteins).

Immunochemical methods

Reproducible immunochemical methods have recently become available for peanut proteins and to a lesser extent for soy proteins. They have the advantage of high sensitivity and of being able to target specifically the proteins of interest (i.e. the identified allergens). However, until the limits of variability of allergen preparations can be established, they are best used as indicators of the presence of allergenic protein, rather than as accurate measures of it. The reason for this is that the accurate measurement of such protein requires that the ratio of different allergenic proteins in the unknown sample mirrors that in the standard. Robustness of the methodology, with respect to interfering substances in the matrix, also needs to be demonstrated. Furthermore, since these immunochemical methods can only be operated in aqueous environments, the assay results will also depend on extraction efficiency.

Conclusions from laboratory experience with specific peanut allergen ELISA kits

- The Prolab, Cortecs and TNO ELISA methods for peanut allergens were compared in Unilever's laboratories where the TNO method was found to be easier to apply and more sensitive than the Prolab one.
- Use of the TNO allergen kit for Ara h1 in Unilever's laboratories showed that most of that particular allergen was recovered in the first extract ($>80\%$).

Vegetable oil refining

Two major routes of oil refining are used commercially, designated chemical and physical refining, respectively.

The refined peanut oil used in the Southampton study on the allergenicity of crude and refined peanut oils (Hourihane *et al.*, 1997a) was prepared by Anglia Oils as outlined below. The process is typical of a chemical refining method.

- *Acid pre-treatment*: oil at 95°C is treated with phosphoric acid (0.12%—final concentration).
- *Neutralization*: sodium hydroxide (17 M diluted to 2.5 M) is added at a concentration equivalent to the free fatty acid concentration + 15%. Soap is removed by centrifugation.
- *Bleaching*: oil at 95°C is treated with bleaching earth (1%) for 25 to 30 minutes.
- *Deodorization*: the oil is heated to 230°C for a minimum of 45 minutes.
- *Typically*, physical refining is conducted as follows:
- *Drying*: oil is dried under vacuum at 95°C.
- *Acid treatment*: oil is treated with citric acid and stirred. Water is then added and the oil stirred.
- *Bleaching*: bleaching earth is added and the oil is stirred.
- *Drying*: oil is dried under vacuum, while being stirred
- *Filtration*: oil is filtered
- *Deodorization*: the oil is heated to 230–250°C for 50 minutes.

Few data exist on the effect of different methods of refining on total residual protein content. While it has been clearly established that refining removes or considerably decreases the allergenicity of oils, the effect of the process in its different forms on the allergenicity of the residual proteins *per se* has not been studied to date. The published literature on oil allergenicity contains few details of the refining process. Unpublished data on the protein content of physically refined oils, albeit sunflower and coconut rather than peanut, determined in a study conducted at the Leatherhead Food Research Association, are summarized in Table 2.

Published data on protein content of vegetable oils

It is accepted that the allergenicity of edible oils, as discussed in this review, is a function of the residual protein remaining after pressing and, if applicable, refining and other processing. The protein content of edible oils has not been investigated systematically and does not currently form part of the product specification. Published values vary widely, depending on the type and source of the oil as well as the methodology used for extraction and analysis. Published results are summarized in Table 1 for the major edible oils for which data are available.

The results indicate generally low levels of protein in peanut oil, whether refined or partly refined. However, the oils designated as crude or unrefined, range from the peanut oil used by Hourihane *et al.* (1997a), which was crude as defined by the refiners and not on general sale, to partly unrefined 'gourmet oils'. Soybean oil also shows generally low levels, except for some of the results by Porras *et al.* (1985).

Recent experimental results on protein content of vegetable oils

Recent experimental results of experiments looking at oils at different stages of refining are summarized in Table 2 (all unpublished data).

Proteins were extracted from the oils by the method of Hoffman (0.2 M ammonium bicarbonate,

Table 1.

Oil	Type	Recovery (%)	Method	Protein (μg/ml)	Reference
Peanut	Alkali-refined, bleached and deodorized	NE	Fractionation of oil and N estimation (Dumas method)	ND	Tattrie and Yaguchi, 1973
	Unrefined/gourmet	NE	Ammonium bicarbonate extraction/ BCA	(10.5)	Teuber *et al.*, 1997a
	Refined	NE	No extraction/RAST inhibition	ND	Nordlee *et al.*, 1981
	Refined (hot-pressed)	60	Ammonium bicarbonate extraction/immunoblotting	ND	Hoffman and Colin-Williams, 1994
	Cold pressed	NE	Ammonium bicarbonate extraction/immunoblotting	0.2–3.3	Hoffman and Colin-Williams, 1994
	Refined	NE	PBS extraction/dialysis vs ammonium bicarbonate/RAST inhibition	ND	Keating *et al.*, 1990
	Refined/peanuts fried	NE	PBS extraction/dialysis vs ammonium bicarbonate/RAST inhibition	equ. 0.05–0.5% peanut	Keating *et al.*, 1990
	Refined/peanuts fried/filtered and steam-cleaned	NE	PBS extraction/dialysis vs ammonium bicarbonate/RAST inhibition	equ. <0.05% peanut	Keating *et al.*, 1990
	Refined	10	PBS extraction/Bradford dye binding	0.12–0.58	Klurfeld and Kritchevsky, 1987
	Crude	NE	Pet. Ether/NaHCO3/BCA	3.4	Olszewski *et al.*, 1998
	Refined	NE	Pet. Ether/NaHCO3/BCA	0.1–0.2	Olszewski *et al.*, 1998
	Unspecified	NE	Not specified	<0.02	Foucard *et al.*, 1997
	Refined	NE	liq N_2, defatting, Rocket immunoeletrophoresis	<0.02	Holzhauser *et al.*, 1998
Soy	Alkali-refined, bleached	NE	Fractionation of oil and N estimation (Dumas method)	0.96	Tattrie and Yaguchi, 1973
	Alkali-refined, bleached and deodorized	NE	Fractionation of oil and N estimation (Dumas method)	ND	Tattrie and Yaguchi, 1973
	Crude	10	PBS extraction/Bradford dye binding	1.9	Klurfeld and Kritchevsky, 1987
	Refined	10	PBS extraction/Bradford dye binding	0.72	Klurfeld and Kritchevsky, 1987
	Unspecified (three samples)	NE	PBS extraction/Inhibition ELISA	110–3300	Porras *et al.*, 1985
	Unspecified (five samples)	NE	PBS extraction/Inhibition ELISA	ND	Porras *et al.*, 1985
	Unspecified, prob. refined	NE	Not specified	<0.02	Foucard *et al.*, 1997
Sunflower	Refined	10	PBS extraction/Bradford dye binding	0.85	Klurfeld and Kritchevsky, 1987

ND = not detected; NE = not evaluated.

Table 2.

Oil	Type	Protein (μg/ml)
Peanut	Crude, Anglia (Southampton)	220
	Refined NBD, Anglia (Southampton)	48
	Crude (Leatherhead[1])	187
	Alkali refined, neutralised and washed (Leatherhead[1])	60
	Refined as above and bleached (Leatherhead[1])	15
	Refined as above and deodorized (Leatherhead[1])	2.2
	Crude (Vlaardingen)	91
	Crude (Vlaardingen) (Total N by DUMAS method)	155
Sunflower	Crude	90
	Super-degummed	57
	As above and bleached	32
	As above and deodorized	0.2
Coconut	Crude	251
	Bleached and filtered	144
	As above and deodorized	7.9

[1]Skinner and Haynes (1998).

48 hr) and analysed using the Pierce Micro BCA method (Smith *et al*., 1985) except where indicated. The Leatherhead analyses were also done using the Lowry assay. Means of the Leatherhead results include both sets of figures. Unilever Research, Vlaardingen used a modification of the Hoffman extraction method: 1 hour at 40°C in phosphate buffered saline + 0.1% Tween.

The recent experimental results show consistent values of the order of 100–300 μg protein per ml of oil for crude oils from both peanut and sunflower. They also reveal that refining results in up to more than 100-fold reductions in protein content (at the final stage) and that the different refining steps all contribute to this reduction both for chemical and physical refining. The final levels in refined peanut oil are about one order of magnitude higher than published results, where these were measurable.

Discussion

Any manifestation of allergenicity due to ingestion of edible oils is due to residual protein. Thus, the safety of such oils for allergic individuals requires demonstration that protein is present at residual levels which are known not to trigger a reaction. Peanut oil is the edible oil that has been most extensively studied, because of the prevalence and severity of peanut allergy.

A consensus exists that crude (unrefined or partially refined) peanut oil can precipitate reactions in peanut-allergic individuals. However, evidence for the allergenicity of refined peanut oil is conflicting, with two distinct positions. The French group of Moneret-Vautrin has produced evidence that some types of peanut oil, stated to be refined oil, may provoke reactions in peanut-sensitive individuals (Moneret-Vautrin *et al*., 1991, 1998). Analytical data on the oils used in one of their studies indicated very low protein contents (100–200 ng/ml for refined and 3.4 μg/ml for crude oil) (Olszewski *et al*., 1998). However, no information was given on efficiency of extraction. In contrast to these studies, the work of Taylor *et al*. (1981) and of Hourihane *et al*. (1997a) showed that the refined peanut oils they used, also typical of the oils available commercially in the USA and UK, respectively, did not provoke any reactions in peanut-sensitive individuals. The two sets of studies differed in several important respects. Moneret-Vautrin's work (Olszewski *et al*., 1998) was a report on a series of clinical cases, whereas both Taylor's and Hourihane's investigations were designed to address the specific question of whether refined oil was safe for peanut allergic individuals. Taylor's study was too small to answer the question convincingly, although the results were clear-cut. However, Hourihane *et al*. employed a sufficient number of volunteers to provide a substantial degree of confidence. Nevertheless, the answers from such trials can never exclude the possibility that extremely sensitive individuals exist who could react to the amount of protein in refined oil, particularly since these individuals may be excluded from the studies for ethical reasons. They can, however, provide an indication of the upper limit of such numbers (5% of the peanut-allergic population in the case of the Hourihane study). While the oils used by Moneret-Vautrin were reported to contain very low amounts of protein (indeed much lower than those measured in the oils used by Hourihane *et al*.), nothing is known about how they were refined. Unpublished evidence from Hourihane *et al*. suggests that the refining process could affect the allergenicity, as well as the quantity of the residual protein. Furthermore, analytical methodology in this field is not standardized, and this makes it difficult to know whether the figures cited in each case are equivalent. A scrutiny of the results of other published studies in this area would suggest that they may not be.

Allergic reactions are dose-dependent phenomena. Thus, thresholds exist below which effects do not occur. Very few studies have been reported into such thresholds. However, a recent investigation by Hourihane *et al*. (1997b) showed that two out of 14

highly sensitive peanut-allergic individuals reacted to 100 μg of peanut protein in peanut flour. No subject reacted to 10 or 50 μg. The reactions observed at the threshold were mild.

Few data are available on the allergenicity of oils made from other oilseeds and those that do exist attest to their safety for allergic individuals. Peanuts are considered to be among the most potent allergens. It is therefore reasonable to assume that, provided the protein content of other edible oils can be shown to be similar to that of peanut oil, then conclusions drawn about the safety of refined peanut oil can be extrapolated to other oils.

Conclusions

The following conclusions can be drawn from the available information on the allergenicity of edible oils and measurements of protein content:

- Soundly designed studies show that chemically refined peanut oil is safe for the overwhelming majority of peanut allergic individuals.
- Confidence in the safety of refined peanut oil is further increased by the demonstration that even highly allergic individuals did not respond to 10 or 50 μg of peanut protein in peanut flour, and only two out of 14 tested responded to 100 μg. Those two reactions were mild and subjective.
- As peanut is acknowledged to be one of the most potent food allergens, it is reasonable to extrapolate the conclusions drawn up for peanut oil to other edible oils.
- Validated analytical methodology for establishing the protein content of oils is required. Once established such methodology should be used to determine the effect of different refining steps on the removal of protein and resulting changes in allergenicity. Data from these experiments would provide valuable data to help improve the safety of edible oils.

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